CHARACTERIZATION OF CLANDESTINE ONE POT METHAMPHETAMINE LABORATORIES USING GAS CHROMATOGRAPHY-MASS SPECTROMETRY AND LIQUID CHROMATOGRAPHY-TANDEM MASS SPECTROMETRY

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Abstract: Since first being seized in 2007, One Pot meth labs have quickly grown to become the number one route of methamphetamine production across the United States. While One Pot meth labs account for 98% of all clandestine labs in the United States, they have remained uncharacterized until now. This research sought to help assess the public health impacts of these clandestine laboratories and assist in criminal investigations by characterizing One Pot meth labs and identifying by-products produced during the illicit manufacturing of methamphetamine. Using gas chromatography-mass spectrometry (GC-MS), by-products were identified in the solid waste (sludge), the liquid waste (post-salt solvent), and the product salts (powdered methamphetamine) of One Pot methamphetamine cooks. Once these by-products were identified, liquid chromatography-tandem mass spectrometry (LC-MS/MS) was used to quantitate the concentration of these by-products in each of the three One Pot meth cook fractions for two organic solvents, namely camp fuel and starter fluid. It was determined that different by-products could be found in each fraction of a One Pot meth cook and that the amount of by-products produced during a cook were affected by the type of organic solvent used during a One Pot meth cook.

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CHAPTER I

INTRODUCTION

Since first being seized in 2007, One pot meth labs have quickly grown to become the number one method of methamphetamine production across the United States.[1] While One Pot meth labs account for 98% of all clandestine labs in the United States, they are currently uncharacterized.[2] It is currently unknown which chemicals are present, and in what quantities, as One Pot meth labs are not yet characterized. This characterization is required to determine potential harmful effects on exposed individuals and the environment.

Although the One Pot method of methamphetamine production remains uncharacterized, other synthesis routes have been characterized. The Red-P and the Birch reduction method, both of which chemically reduce ephedrine or pseudoephedrine to methamphetamine as the One Pot method does, have been well characterized, and the by-products produced by each method have been documented.[3,4] The United States Environmental Protection Agency (EPA) has taken advantage of the characterization of these cooks and has published warnings about the types of cleaning chemicals that should and should not be used to clean these types of meth labs. In addition, the EPA mentions that certain cleaners may react to by-products created during meth cooks, although the

reactivity is unknown, as the by-products themselves are unknown. Because of the lack of knowledge about the by-products produced, the EPA does not suggest the use of harsh cleaning chemicals that may react with these by-products until the by-products are properly identified and research is performed on how they interact with the cleaning chemicals.[5]

Identifying by-products associated with the One Pot method of methamphetamine production would have several benefits. Besides allowing for research to be done on how certain cleaning chemicals react with the by-products, thus allowing better guidance for meth lab clean-up crews on how to remediate former meth labs, by-product identification would also enhance the safety of meth lab first responders and clean-up crews. Currently, how well or how quickly meth-lab related chemicals can permeate different materials is unknown.[5] The identification of by-products of One Pot meth labs allows for testing on the suits worn by first responders and clean-up crews to determine the length of time before the by-products breach their protective clothing. This testing would also allow for further research into what health effects these by-products may cause. Identification of One Pot meth lab by-products also offers law enforcement a tool to try and uncover these clandestine labs. If a by-product is unique to One Pot methamphetamine manufacturing, that by-product may be able to be traced through the air or through the sewers to the location of a One Pot meth lab.[6]

The purpose of this study was to test the theory of by-product identification in clandestine One Pot methamphetamine labs. Methamphetamine was synthesized at the Oklahoma State University Forensic Toxicology and Trace Laboratory (OSU-FTTL), using two commonly encountered street solvents, starter fluid (diethyl ether) and camp

fuel (light petroleum distillate). Each meth cook was comprised of three components: the liquid waste (post-salt solvent), the solid waste (sludge), and the products (product salts). All components were analyzed for the presence of unique chemical by-products using gas chromatography-mass spectrometry (GC-MS) and liquid chromatography-tandem mass spectrometry (LC-MS/MS).

Once the One Pot cooks had been analyzed with GC-MS and LC-MS/MS, the data was analyzed in relation to the following research questions: 1.) Does waste generated from a One Pot methamphetamine lab contain chemical by-products unique to this cook method? 2.) Are any by-products observed in One Pot methamphetamine waste currently unidentified compounds? 3.) Does the type of solvent used during a One Pot methamphetamine cook make a difference in the by-products observed in the cook waste? The answers to these questions will provide law enforcement agencies specific target compounds to aid in the discovery and dismantling of clandestine One Pot methamphetamine labs, as well as to aid meth lab clean-up crews in ensuring previously contaminated sites are safe for human occupancy.

CHAPTER II

REVIEW OF LITERATURE

2.1 Introduction

Methamphetamine, or meth, is a stimulant that increases the release of the neurotransmitter dopamine, while also blocking its reuptake, leading users to feel the energizing effects of a high for prolonged periods of time. The prolonged high associated with methamphetamine is what makes it so addictive, causing many to dub meth as the most addictive drug in the world.[7] In 2012, the National Institute on Drug Abuse estimated that there were over 12 million methamphetamine users worldwide, accounting for more drug users than cocaine and heroin combined.[7,8] In 2014, the United States Drug Enforcement Administration (DEA) reported 9338 clandestine, or secret and illicit, meth labs; these were only the labs that were discovered.[9,10] Many more clandestine meth labs are suspected of being operational, with the method of methamphetamine production varying from lab to lab.

Over the course of history, meth has been synthesized, or "cooked," by a variety of different methods, including the "P2P" method, the "Red-P" method, the "Nazi" method, and the "One Pot" method.[11] As the US government identified these production methods, laws to regulate the sale of precursor products were put into effect, causing the meth cooks to develop new ways to produce their products. Currently, the method of

choice for methamphetamine production is the One Pot, or shake-and-bake, method, which allows cooks to produce meth quickly with a small quantity of starting materials, all of which can be easily obtained in local stores. Not only are the materials easy to obtain, but these cooks also can be done in something as simple as a plastic bottle, making these meth "labs" mobile and easy to hide.[11]

Due to the ease of obtaining these starting materials and the ability to easily conceal the labs, One Pot cooks have increased in prevalence across the United States, causing law enforcement agencies to begin searching for a better way to detect these clandestine labs.[4] The identification of these labs has proven difficult for law enforcement agencies, as no information is currently available about what chemicals besides methamphetamine are produced during a One Pot meth cook. Without knowing what other chemicals are produced in a One Pot cook, law enforcement agencies are limited in what they can look for to positively identify the presence of a meth lab. To identify chemical by-products that are characteristic to a One Pot methamphetamine cook, such a cook must be performed and characterized in a laboratory setting. This type of chemical characterization process has not yet been done for the One Pot method of methamphetamine production.

2.2 History

Methamphetamine was first synthesized in 1893 by the Japanese pharmacologist Nagayoshi Nagai. Nagai produced methamphetamine by chemically reducing ephedrine extracted from the plant family *Ephedra*. In 1919, Akira Ogata further purified methamphetamine, becoming the first person to synthesize it in the crystalline form,

known as crystal meth, which is a purer form of the drug that has the allure of being smoked.[12]

During World War II, methamphetamine was used to keep soldiers up for days at a time. The Japanese even gave their Kamikaze pilots meth before they embarked on suicide missions. When World War II ended, the surplus methamphetamine from the military was sold to the public, leading to an epidemic of methamphetamine use. In the post-World War II era, methamphetamine was used for its ability to suppress appetites and to increase energy and focus, causing a variety of people from college students to overworked factory employees to seek the drug for help in everyday life.[13]

During the 1960s, intravenous use of methamphetamine began to increase; the drug culture wanted strong drugs that would take effect quickly. The pursuit of strong, fast-acting drugs led to a greater amount of intravenous drug use, which allows the effects of drugs to be felt almost instantaneously. Due to the high water solubility of methamphetamine, it quickly became a heavily used intravenous drug until 1970, when meth was finally outlawed in the United States by the Controlled Substance Act.[13,14]

After being classified as a Schedule II controlled substance, meaning the substance has a high potential for abuse and dependence yet is accepted for medicinal use, methamphetamine began being produced and distributed by motorcycle gangs.[13,14] Known by the street name "poor man's cocaine," meth was mostly sold to poor, rural Americans who could not afford other stimulants, such as cocaine. During this period of motorcycle gang run illicit methamphetamine drug markets, clandestine meth labs began popping up across the United States.

In the 1990s, Mexican cartels began trying their hands in the production and distribution of methamphetamine. Large Mexican meth labs could produce approximately 25 pounds of highly pure methamphetamine each day.[13] Due to its vast availability and high purity, many people have resorted to buying their meth from the Mexican cartels, but a substantial number of people still find that producing their own methamphetamine is easier and cheaper.[15]

2.3 Manufacturing Methods

Over the years, methamphetamine has been illegally produced in clandestine laboratories using one of the many variations of two unique processes. The first process, known as the P2P method, uses phenyl-2-propanone (P2P) and methylamine to synthesize methamphetamine.[16] Following the DEA's effort to minimize the production of methamphetamine, P2P was classified as a Schedule II controlled substance in 1980; methylamine was also put on the DEA's "watch list" for chemicals known to be precursors to illicit drug production.[14,16] The second process commonly used to produce methamphetamine in clandestine laboratories is the reduction of ephedrine, or pseudoephedrine. The ephedrine reduction process is easier process to perform than the P2P method, making the ephedrine reduction more popular with meth cooks lacking a chemistry background.[17]

The manufacturing method of methamphetamine plays an important role in the potency of the drug. Methamphetamine has two chemical isomers: d-methamphetamine and l-methamphetamine. Chemically, these isomers only differ by the direction the methyl group on the alpha-carbon is facing. Physiologically, these isomers act very differently. Both cause the release of and block the reuptake of dopamine, but d-

methamphetamine does so in a much greater fashion, leading to a stronger and more prolonged high than l-methamphetamine produces. Due to l-methamphetamine's poor ability to cause a high, meth cooks have tried to limit how much is made during their production of d-methamphetamine.

2.3.1 P2P Method

As stated earlier, the P2P method uses phenyl-2-propanone and methylamine, both of which are intermediates for the production of pesticides, as the precursor chemicals for the synthesis of methamphetamine.[18,19] The P2P method of methamphetamine production is a complex method that requires some knowledge of organic chemistry in order to obtain a satisfactory yield of d-methamphetamine, as the method is equally as likely to produce unwanted l-methamphetamine. On top of the need for meth cooks to have knowledge in organic chemistry in order to produce a high yield of methamphetamine, the P2P method is also time-intensive. A single reaction can take as long as 3 days to perform and generally requires several cooks to be present in order to complete. Also, the cooks must have their own laboratory gear to perform the cook, and they must be available to stir the reaction every 30 minutes.[16] In 1980, P2P became classified as a Schedule II controlled substance, and methylamine was placed on the DEA's watch list for known precursors to illicit drug manufacturing. This listing made methamphetamine production with the P2P method difficult. At first, many cooks began synthesizing P2P and methylamine on their own, a process that often resulted in lead contamination of the resulting methamphetamine.[20] As time went on, the better clandestine drug chemists began searching for a way to produce methamphetamine without P2P or methylamine.

2.3.2 Red-P Method

As drug chemists and cooks searched for a way to produce methamphetamine without drawing the attention of law enforcement agencies, they began using commercial chemicals as opposed to buying pure chemicals directly from chemical supply companies. By 1982, experimentation with commercial chemicals in the production of methamphetamine led cooks to discover that ephedrine and pseudoephedrine could be used as precursor chemicals for production. Not only were these chemicals unregulated, but the cook process was also much easier and much faster than the P2P method of production. In addition, the resulting methamphetamine produced from the reduction of ephedrine contained almost exclusively d-methamphetamine, as opposed to the P2P method which produced a 50-50 mixture of l- and d-methamphetamine.[16,20]

Although the DEA busted its first ephedrine- and pseudoephedrine-based clandestine lab in 1987, the ability to reduce ephedrine to methamphetamine was known long before that time.[21] Both ephedrine and pseudoephedrine come from the plant family *Ephedra*, which was being studied by Nagai in 1983 when he first accidently manufactured methamphetamine.[12] Ephedrine and pseudoephedrine have chemical structures very similar to that of methamphetamine and can be easily reduced to produce meth. The ephedrine reduction process was first observed in the United States during a DEA seizure of a meth lab that was producing methamphetamine via the Red-P method.[21]

The Red-P method of methamphetamine synthesis requires the use of red phosphorous and hydroiodic acid to reduce ephedrine to methamphetamine. The ephedrine or pseudoephedrine used in the Red-P method needs to be extracted from over-the-counter cold tablets before the cook can proceed. Once isolated, the ephedrine is

combined with hydroiodic acid. Since hydroiodic acid is a controlled chemical, many cooks have made it during the reaction by combining iodine crystals, usually from household disinfectants, and hydrochloric acid. This process has allowed them to continue methamphetamine production without drawing the attention of law enforcement officials. Red phosphorous is also a controlled chemical so cooks generally must obtain it by scraping the striker plate off of matchbooks or by disassembling road flares. The Red-P method of methamphetamine production takes 18-72 hours to perform and produces a 50-75% yield of d-methamphetamine.[22] In other words, every gram of ephedrine put into a cook yields 0.5-0.75 grams of d-methamphetamine.

2.3.3 Birch Reduction Method

With the introduction of the Comprehensive Methamphetamine Control Act of 1996, iodine became difficult to obtain in the quantities needed to perform the Red-P method of methamphetamine production.[23] This difficulty in obtaining precursor chemicals once again led drug chemists to search for a new way to synthesize methamphetamine. The result was the Birch reduction method of methamphetamine production.

The Birch reduction, or "Nazi," method combines ephedrine, anhydrous ammonia and lithium in some sort of flat baking dish. The lithium is dissolved by the ammonia, freeing electrons, which can then be used to reduce ephedrine or pseudoephedrine to methamphetamine. This method of methamphetamine production became popular with cooks, as all of the precursors were easily obtained commercial goods with the exception of anhydrous ammonia; this precursor could be stolen or bought from farmers with relative ease.[24] While the Comprehensive Methamphetamine Control Act of 1996 did

restrict the import of bulk shipments of ephedrine products, meth cooks were able to obtain enough ephedrine from over-the-counter cold medications to enable them to produce methamphetamine.[23] Another benefit of the Birch reduction method is its ease and its short cook time. To produce methamphetamine with the Birch reduction method, the ingredients listed above are simply added to a container in a specific order, mixed, and allowed to react for 6-9 hours.[25] After the reaction is complete, the liquid portion is blasted with hydrogen chloride gas, made by combining rock salt and sulfuric acid in an enclosed container; hydrogen chloride gas causes the methamphetamine to precipitate out of solution. The resulting methamphetamine is mostly d-methamphetamine if the cook has been careful to use over-the-counter cold medications that contain either l-ephedrine or d-pseudoephedrine.[21]

As word of this new, quicker method of methamphetamine production reached law enforcement agencies, there was a push to put tighter regulations on the precursor chemicals. Since the sale and storage of anhydrous ammonia was already regulated, legislation focused on regulating the amount of ephedrine and pseudoephedrine purchased in a given amount of time. The Combat Methamphetamine Epidemic Act of 2005 did just that. This act limits an individual to purchasing 9 grams of ephedrine or pseudoephedrine per month. Stores also began requiring a photo identification and a signature when someone wishes to purchase an ephedrine product; this information is entered into an automated system that records the purchase of an ephedrine product and links the purchase to the buyer. The automated system enables store clerks to determine how much ephedrine a person has bought in the last day, week, month, and year, allowing the clerk to deny the sale to anyone who has already reached the purchasing

limit for ephedrine products.[26] Many retail stores also began pulling ephedrine products from their shelves and storing them behind the pharmacy counter in order to combat theft.[20] The benefits of the Combat Methamphetamine Epidemic Act of 2005 were almost immediately noticed, as meth lab incidents decreased from 2005-2007.[24]

As with earlier legislation aimed at the regulation of methamphetamine precursors, meth cooks found a way to get around the limit on ephedrine and pseudoephedrine purchases. Some cooks began hiring "smurfs," who would travel around an area, usually with numerous fake IDs, purchasing the limit of ephedrine products and then selling them back to the meth cooks. Those that did not want to waste the money hiring smurfs began searching for a way to limit their ephedrine needs, leading to the development of the One Pot method for methamphetamine production.[24]

2.3.4 One Pot Method

The One Pot method of methamphetamine production relies on the same chemical reactions as the Birch Reduction method, except instead of the use of liquid anhydrous ammonia, ammonia gas is created when sodium hydroxide and ammonium nitrate are combined in an organic solvent. Unlike the Birch Reduction method, which typically requires over 30g of ephedrine to produce a batch of methamphetamine, the One Pot method can be performed with a single package of cold medication. The chemicals are commonly observed being mixed in a plastic soda bottle, which acts as the reaction vessel and prevents the generated ammonia gas from leaving the reaction. With such a small "lab," meth cooks are able to easily start a One Pot cook in one location and then leave that location with lab concealed in a backpack or purse. On top of being highly concealable, the One Pot method is an easy chemical procedure to perform and can be

completed in as little as two hours. The ease and speed of One Pot meth cooks, as well as the willingness of smurfs to gather ephedrine products for monetary gain, led to an increase in meth lab incidents beginning in 2008, with a majority of these incidents being related to meth cooks using the One Pot method to synthesize small batches of methamphetamine for their own personal use.[24]

2.4 Impact of Meth Labs

The prevalence of clandestine One Pot methamphetamine labs is on the rise. These labs are easy to hide and contain many dangers. Many of the cooks that operate meth labs have no idea what they are doing; they are simply following a recipe given to them by another cook. Because of this lack of knowledge, many cooks end up either mixing together chemicals that should not be mixed or not letting reactions finish completely. This uneducated cook process causes many unknown compounds to be created and a significant amount of hazardous waste to be produced, leading to an extremely unsafe environment for anyone who discovers or cleans up sites of methamphetamine production.[27]

Currently there is no national requirements for the cleanup of clandestine methamphetamine laboratory sites once they have been dismantled by law enforcement. The EPA has laid out guidelines that include testing the level of known hazardous chemicals before cleanup, airing out the lab site, vacuuming carpeted areas, flushing out plumbing and heat/air ducts, washing walls/ceilings/hard flooring with detergents, and then retesting the site for hazardous chemicals.[5] While this plan shows how levels of known chemicals decrease after cleanup, some chemical hazards that are produced by

meth cooks are unknown, thus the chemicals cannot be tested for and may still be present after cleanup efforts have ceased.

With the presence of unknown chemicals in prior meth labs, the cleanup procedure itself can be highly dangerous. When cleaning these labs, workers need to be cautious about what type of cleaners they use. Some cleaners can react with residual chemical contamination, causing unwanted reactions that may produce hazardous gases, such as the reaction that happens when bleach is combined with sulfuric acid.[28] When these types of reactions are known, cleanup crews can use caution to avoid cleaners that may cause such a reaction to occur. When the residual by-products from a meth lab are unknown, as is the case for the One Pot method, cleanup crews enter the lab with very little knowledge of what chemicals they are cleaning up or how to clean them up safely.

Due to the lack of knowledge about all of the by-products formed during the One Pot production, the long-term health effects of being exposed to clandestine meth labs is also unknown, and studies about these effects are sparse. Currently it is even unclear if the personal protective equipment being used by law enforcement and meth lab cleanup crews can filter out all of the unknown compounds found in a meth lab.[27] The unknown by-products of One Pot meth labs can also affect many more people than those who have been directly exposed to the lab. For every pound of methamphetamine produced in a clandestine One Pot laboratory, 5-7 pounds of toxic waste are produced.[29] The waste produced is often disposed of in the environment in some way, such as by being thrown in the trash, dumped in a ditch, or poured down the drain. Once released into the environment, methamphetamine waste can make its way into the wastewater system and eventually even into the drinking water supply. While wastewater treatment does

eliminate some of the toxic waste, research has shown that 1-48% of amphetamine-like chemicals that enter the wastewater system are still present in the post-treatment water.[30] The environmental release of methamphetamine waste is a major health concern, as recent analyses of wastewater show increased amounts of illicit and pharmaceutical drugs in the system. With the increase in meth lab prevalence around the country, more waste is being generated and introduced into the waste water system every year. The long-term effects of these meth lab waste products are still unknown.[31]

2.5 Current Meth Lab Characterizations

Currently, much effort is being put into finding a rapid analytical test that can be used to identify methamphetamine in clandestine labs during cleanup efforts. Liquids found in such meth labs can be difficult to analyze and are normally discovered in unmarked containers. Many of the people who run such labs have no chemistry knowledge, causing these cooks to stop reactions before they come to completion or allowing the reactions to continue longer than needed. This varied reaction time leads to the liquids in question containing not only the desired product, methamphetamine, but also many by-products, which are produced due to excess starting materials, over-reacted products, and reaction intermediates formed during the cook.[32] To add to the difficulty in identifying what is in the unknown liquids found in a clandestine meth lab, many labs examined today are One Pot meth labs, which have not yet been characterized, thus the by-products that are formed are currently unknown.

While the by-products of One Pot methamphetamine lab are unknown, much research has been put into the identification of by-products of other methods of methamphetamine production, such as the Birch reduction method of methamphetamine

synthesis.[4] As stated earlier, Birch reduction labs rely on similar chemistry as the One Pot labs, using a metal in the presence of ammonia to reduce ephedrine or pseudoephedrine to methamphetamine. Due to the similar chemistry of the Birch reduction and One Pot methods, current assumptions are that the by-products produced by these two methods are the same. Previously research has observed only one major by-product in Birch reduction labs, 1-(1,4-cyclohexadienyl)-2-methylaminopropane (CMP).[33,34] One Pot meth labs remain uncharacterized.

In 2012, new research emerged about the by-products formed during the Birch reduction method of methamphetamine production. Contrary to previous research that listed CMP as the only by-product in this method of methamphetamine production, Kunalan, Kerr, and Daéid reported the discovery of multiple Birch reduction by-products, including one compound whose identity is currently unknown. The researchers also observed that by slightly differing the starting material used in a Birch reduction cook, such as using the freebase form of ephedrine instead of the salt form, different by-products were produced.[4] This recent contradiction to previously published material raises questions about what other by-products of different methamphetamine production methods remain undocumented and, of those, how many have had their chemical structure identified.

As with the Birch reduction method, the Red-P method of methamphetamine synthesis has a plethora of by-products associated with it. Small changes in the amount of precursors added, the order of precursor additions to the reaction, the time the reaction was allowed to reflux, and the type of precursors used all affected the by-products observed in this method of meth production.[3]

Recently, there has been a greater focus on using ambient mass spectrometry to analyze meth cooks in real time, with Birch reduction and Red-P cooks receiving a majority of this focus.[35] These cook methods have been a focus because the Birch reduction method is currently the most popular method in the United States for the mass production of methamphetamine and, while no longer highly used in the United States, the Red-P method is still the primary method of methamphetamine production in Asia.[3,36] The use of ambient mass spectrometry allows researchers to examine these cook methods at specific steps during the reaction, enabling for the identification of certain key chemicals that are present at each stage of the reaction, such as the ratio of ephedrine to methamphetamine during each step of a Birch reduction or a Red-P meth cook.[35] Even with the extreme popularity of One Pot meth cooks throughout the United States, analytical characterization has not been performed on a One Pot meth cook. Lack of characterization research is likely due to the One Pot method's inability to produce large quantities of methamphetamine in a single cook.

2.6 Conclusion

As shown above, methamphetamine is a major problem in the United States. Older routes of methamphetamine synthesis have been combated by the introduction of several pieces of legislation. This legislation has focused on limiting the availability of several precursor chemicals that are required for methamphetamine synthesis, including, but not limited to, anhydrous ammonia, phenyl-2-propanone, ephedrine, and pseudoephedrine.

Just as law enforcement has adopted laws to limit methamphetamine precursors, so too have meth cooks adapted their cook processes to skirt around newly adopted laws.

Currently, methamphetamine cooks are predominantly using the One Pot method to produce their products quickly and discreetly. The mobility of these labs is allowing cooks to produce methamphetamine practically anywhere, including in cars, houses, and even personal backpacks.[37]

Seized clandestine meth labs provide hazards not only to those directly involved in their cleanup but also the public as a whole. With One Pot meth cooks currently uncharacterized, the identity of chemical by-products that may be present during and after cleanup procedures, as well as by-products that are released into the environment once a One Pot meth lab is discarded, are unknown. The Birch reduction method of methamphetamine synthesis, which causes the precursor chemicals to undergo a similar series of chemical reactions as the One Pot method, can provide an idea of what types of by-products may be formed during a One Pot meth cook; however until a One Pot cook is formally characterized, these by-products remain unknown.[38]

The focus of this research is to characterize the steps of a One Pot methamphetamine cook and identify unique chemical markers that can signal the production of methamphetamine. Methamphetamine will be produced via the One Pot method in a laboratory setting and then all parts of the One Pot cook, including the liquid waste, the solid waste, and the methamphetamine itself, will be analyzed to identify any unique chemicals produced during the cook. This characterization will be done with gas chromatography-mass spectroscopy (GC-MS) and liquid chromatography-tandem mass spectroscopy (LC-MS/MS). This research aims to answer whether or not the One Pot method of methamphetamine production leads to the formation of unique chemical by-

products that could be used to better identify clandestine One Pot meth labs and to aid cleanup crews in safely removing hazards associated with a seized One Pot lab.

CHAPTER III

METHODOLOGY

3.1 Introduction

This research project was a two-part study: production of methamphetamine and laboratory analysis. The production of methamphetamine was performed in the Oklahoma State University Forensic Toxicology and Trace Lab (OSU-FTTL). For the second part of the study, the One Pot meth cook was split into three components, all of which were analyzed with gas chromatography-mass spectrometry (GC-MS) and liquid chromatography-tandem mass spectrometry (LC-MS/MS). The three meth cook components were the solid waste (sludge), the liquid waste (post-salt solvent), and the final product (product salts). All three components were analyzed for the presence of precursors, products, and unique by-products. To the knowledge of the research group, identification of by-products unique to the One Pot method of methamphetamine production have not been characterized prior to this research. The purpose of this research was to identify said by-products and determine if they were unique to the One Pot method of methamphetamine production. The research performed did not involve human specimens so Institutional Review Board (IRB) regulations and guidelines will not be mentioned. OSU-FTTL is a DEA registered entity for Schedule I-V controlled substances (methamphetamine is Schedule II). While the ingredients for the One Pot

meth cooks used during this research are listed, no amounts are given to avoid illegal use of the method.

3.2 Methamphetamine Synthesis

3.2.1 Materials

All reagents and materials except for ammonium nitrate, pseudoephedrine-HCl/ephedrine-HCl tablets, and Nanopure water were purchased from commercial suppliers. To simulate a street cook, ammonium nitrate was obtained from instant cold compress packs (GoGoods.com, Inc, Columbia, MD); Coleman®* Camp Fuel (Model: 5103B253 Coleman®, Wichita KS), from a local hardware store; and a ground mixture of pseudoephedrine-HCl and ephedrine-HCl tablets, from a government source. ACS grade sodium hydroxide beads, hydrochloric acid (37%), ACS grade diethyl ether to simulate starting fluid, and dichloromethane were purchased from VWR Analytical (VWR, Sugar Land, TX). Lithium ribbon and 99^{+}% hydrogen chloride gas were purchased from Sigma (Sigma-Aldrich Corp, St. Louis, MO). Ammonium formate was purchased from Alfa Aesar (Alfa Aesar, Ward Hill, MA). Formic acid was purchased from EDM (EDM Millipore Corp, Billerica, MA). Methanol was purchased from JT Baker (Avantor Performance Materials Inc, Center Valley, PA). Nanopure water was obtained through the use of a Barnstead Nanopure Diamond laboratory water system (Thermo Scientific, Waltham, MA).

Amphetamine, Amphetamine-d_6, Methamphetamine, Methamphetamine-d_5, 1S,2S(+)-Pseudoephedrine, and 1S,2R(+)-Ephedrine-HCl standards were all purchased at

* Coleman® Camp Fuel was used during this research due to its availability. This is not meant to imply that Coleman® is the only camp fuel (light petroleum distillate) used in the illicit production of methamphetamine or endorse its use.

a concentration of 1 mg/mL from Cerilliant (Cerilliant Corp, Round Rock, TX). Pseudoephedrine-d_3 HCl and 1S,2R(+)-Ephedrine-d_3 HCl standards were also bought from Cerilliant at a concentration of 100 µg/mL. CMP-HCl standard was purchased at a concentration of 1 mg/mL from Cayman (Cayman Chemical, Ann Arbor, MI).

3.2.2 Sample Production and Collection

The One Pot cooks in this study were performed using identical reaction materials, except for use of 2 different organic solvents as described below. The cook procedure employed was previously developed at Oklahoma State University Center for Health Sciences as a modification of a street lab recipe. Six identical cooks were performed over the span of 3 days, differing only in selection of solvent: 3 cooks used laboratory-grade diethyl ether (ether) and 3 used camp fuel. On each of the 3 cook days, an ether cook was performed alongside a camp fuel cook to help reduce variability between the 2 cooks. Diethyl ether cooks are designated E1, E2, and E3 in this report; camp fuel cooks are designated C1, C2, and C3. All chemicals were weighed prior to the reaction with a measured precision of less than 0.1 g difference per weighed ingredient between the 2 cook types.

For each cook, 1 g of ground pseudoephedrine-HCl/ephedrine-HCl tablets was added to 250 mL of solvent, along with a proprietary ratio of Nanopure water, ammonium nitrate, sodium hydroxide, and lithium ribbon, cut into 2 cm pieces. All ingredients were added to a 500 mL pressurized glass-reaction flask, which was then capped. An off-gassing apparatus, consisting of valve tubing rated for 150 psi, was inserted into the lid of the reaction vessel and included a pressure gauge (SSI Technology, Inc, Janesville, WI), an emergency pressure release valve rated for 90 psi,

and a manual blocking valve. Valve tubing coming from the blocking valve was inserted into a receptacle filled with water. The off-gassing apparatus setup is shown in Figure 1. The complete cook setup is shown in Figure 2. Immediately following the start of the reaction, the system was left open to the fume hood atmosphere. After 30 seconds, the manual blocking valve was closed, sealing the system for two hours while the reaction proceeded.

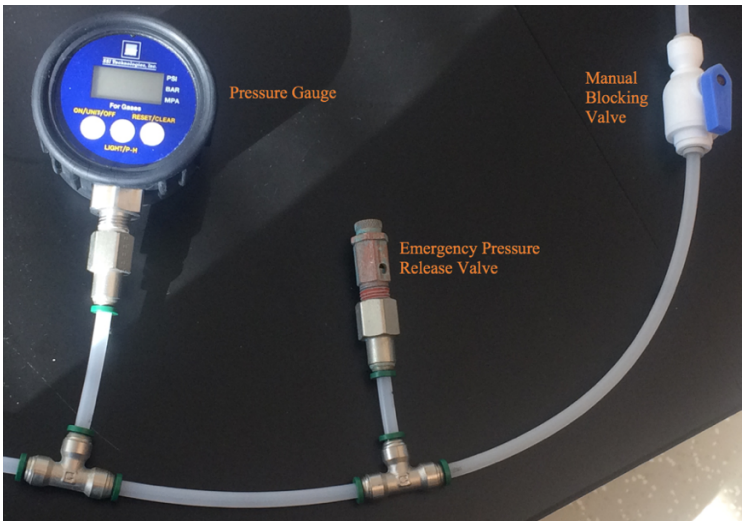

Figure 1. Off-gassing apparatus used during the methamphetamine cooks.

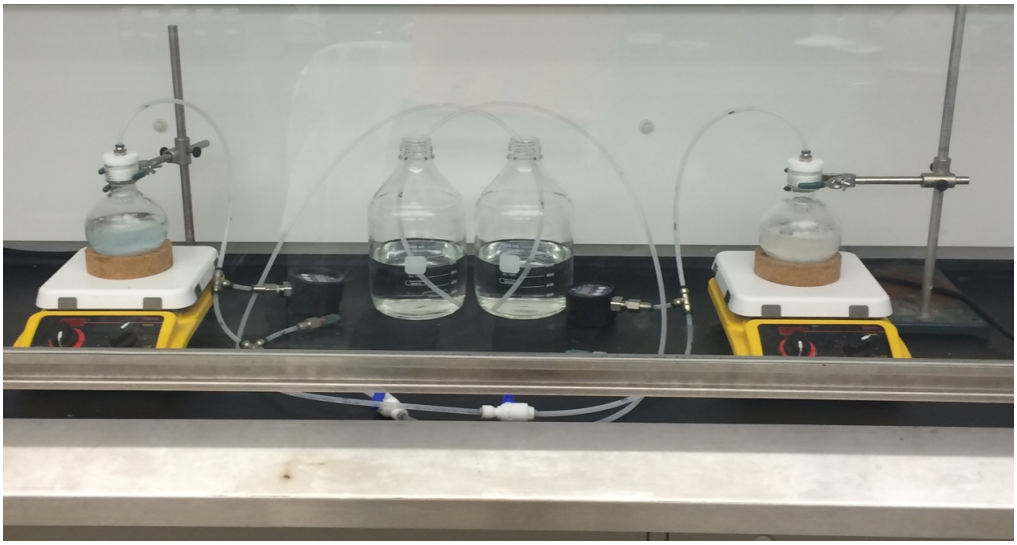

Figure 2. A 2-cook tandem reactor setup containing a camp fuel cook (left reaction vessel) and an ether cook (right reaction vessel), including off-gas apparatus and bubblers.

After 2 hours, the manual blocking valve was opened slowly to bleed off volatiles from the reaction vessel. All reaction gasses were bubbled into a receptacle containing water, which served as a buffer to control the release of ammonia gas into the fume hood. Reaction completion was determined by cessation of visible bubbling of the solvent mixture, at which time the off-gassing apparatus was removed from the cook vessel.

After the off-gassing apparatus was removed, the gasketed round-bottom flask lid was unscrewed and removed. For safety, the lithium was removed and quickly dropped into a container of water to allow the lithium ribbon to finish reacting. Once the lithium was removed from the mixture, the cap was loosely placed on top of the flask to allow any remaining ammonia gas that was produced during the cook to be vented. The flask was then left overnight to completely vent any residual ammonia gas.

After the reaction was vented overnight, mother liquor was filtered using coffee filters (Farmer Bros Co., Ft. Worth, TX) that had been pre-moistened with the solvent of the respective cook. After filtration, the coffee filter was dunked into water so any trace lithium present would react. The sludge was dissolved in 250 mL of Nanopure water and saved for analysis. After collection of the sludge, the filtrate was purged with hydrogen chloride gas from a 227 g lecture bottle to salt out the methamphetamine. Once precipitation visually ceased, the product salt was recovered with another coffee filter, dried, and weighed.

After weighing, a small sample of product salt was subjected to 2 different NIK Public Safety Narcotics Identification System presumptive color tests (NIK Public Safety Inc, Jacksonville, FL) to verify presence of methamphetamine. The first was Test A: Marquis Reagent. The appearance of an orange/brown color indicated the presence of

amphetamines. If the Marquis test was positive, Test U: Methamphetamine or MDMA (Ecstasy) was used. If Test U yielded a dark purple color, the product salts could then be assumed to contain methamphetamine, indicating the subject cook was successful. Examples of positive Marquis and methamphetamine tests can be seen in Figure 3. The product salts were also analyzed with a FirstDefender RMX RX2863 Raman spectrometer (Thermo Scientific, Waltham, MA) to test for the presence of methamphetamine. Figure 4 shows the results obtained from the Raman analysis of the E2 product salt. After presumptive testing, the product salts were secured and the post-salt solvent waste was retained for analysis.

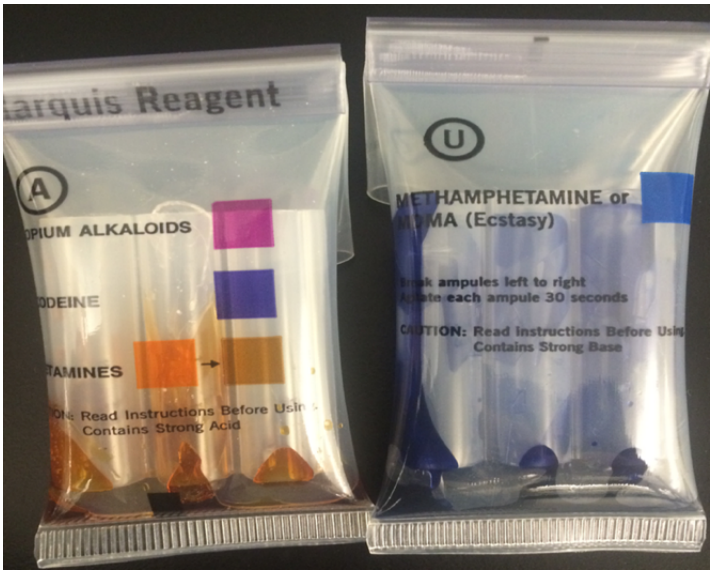

Figure 3. Examples of positive NIK Public Safety Narcotics Identification System presumptive color tests. Test A: Marquis Reagent (left) was used to identify the presence of amphetamines. Test U: Methamphetamine and MDMA (Ecstasy) (right), was used to identify the presence of methamphetamine.

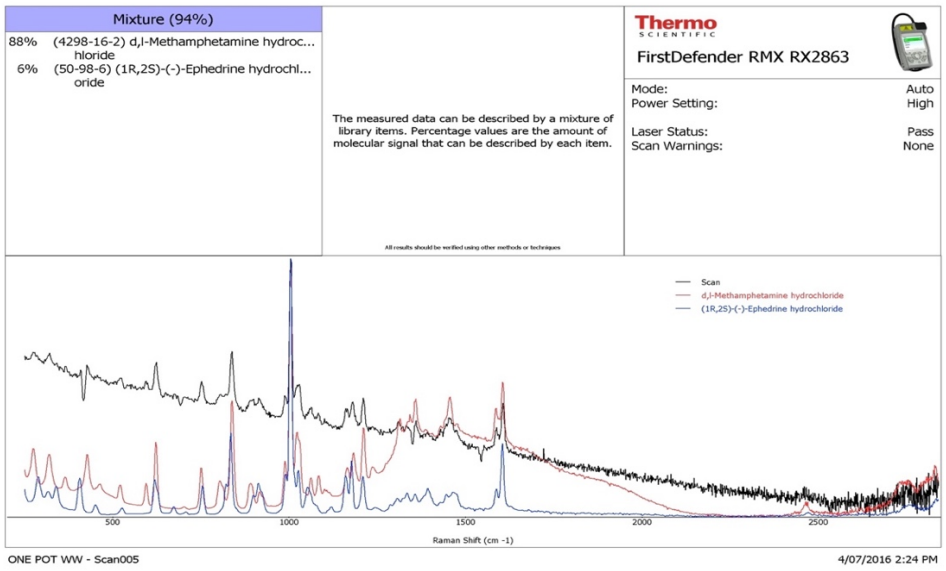

Figure 4. Raman spectrum obtained from analysis of the E2 product salt.

3.3 Sample Preparation

3.3.1 Post-Salt Solvents

The post-salt solvents were extracted via liquid-liquid extraction (LLE) and then analyzed with GC-MS. First, 2000 µL of post-salt solvent was added to a plastic 8 mL test tube and acidified with 100 µL of 37% HCl. This mixture was capped and vortexed at 3000 rpm for 10 seconds. After vortexing, 1000 µL of deionized water was added to the test tube, which was then recapped and vortexed for 15 seconds. The tube was allowed to sit until the organic and aqueous layers separated, at which point the organic layer (Fraction A) was removed and placed into a clean 8mL test tube. Next, 2000 µL of dichloromethane was added to the test tube containing the remaining acidic aqueous layer; 200 µL of concentrated sodium hydroxide was then added to the mixture to create a basic solution. The mixture was vortexed for 15 seconds. The mixture was allowed to sit until the organic (Fraction B) and aqueous layers separated, at which time the aqueous layer was removed. Both organic layers (Fractions A and B) were then dried to complete

dryness under a stream of nitrogen and reconstituted in 100 µL of dichloromethane. The reconstituted solutions were combined to give a total of 200 µL of sample for GC-MS analysis.

For LC-MS/MS analysis, the post-salt solvents were diluted in water at a ratio of 1:10 000. One milliliter of this mixture and 10 µL of internal standard solution was added to a sample vial and analyzed with LC-MS/MS. The internal standard solution used contained 1000 ng/mL of Methamphetamine-d_5, Amphetamine-d_6, Pseudoephedrine-d_3 HCl, and 1S,2R(+)-Ephedrine-d_3 HCl in water.

3.3.2 Sludge

LLE was used to prepare the sludge for GC-MS analysis. During the cooks, the freshly-filtered wet sludge was dissolved in 250 mL of Nanopure water for later wet-chemistry analysis. First, 2000 µL of the sludge-water mixture was added to a plastic 8 mL test tube and made basic with 100 µL of concentrated sodium hydroxide. This mixture was capped and vortexed at 3000 rpm for 10 seconds. After vortexing, 2000 µL of dichloromethane was added to the test tube, which was then recapped and vortexed for 15 seconds. The tube was allowed to sit until the organic (Fraction C) and aqueous layers separated, at which point the aqueous layer was removed and placed into a clean 8 mL test tube. Next, 2000 µL of dichloromethane was added to the test tube containing the aqueous layer; 600 µL of 37% HCl was then added to the mixture to acidify it. The mixture was vortexed for 15 seconds. The mixture was allowed to sit until the organic (Fraction D) and aqueous layers separated, at which time the aqueous layer was removed. Both organic layers (Fractions C and D) were then dried to complete dryness under a

stream of nitrogen and reconstituted in 100 µL of dichloromethane. The reconstituted solutions were combined to give a total of 200 µL of sample for GC-MS analysis.

For LC-MS/MS analysis, the sludge was diluted in water at a ratio of 1:100 000. One milliliter of this mixture and 10 µL of internal standard solution was added to a sample vial and analyzed with LC-MS/MS.

3.3.3 Product Salts

In preparation for GC-MS analysis, all product salts were dissolved in methanol at a concentration of 1 mg/mL. Further dilutions were performed in methanol to concentrations of 100, 50, 10, 1, 0.1, and 0.01 µg/mL, with 50 µg/mL determined to produce the best instrument response.

For LC-MS/MS analysis, the product salts were diluted in water to a final concentration of 100 ng/mL. One milliliter of the 100 ng/mL product salt mixture and 10 µL of internal standard solution was added to a sample vial and analyzed with LC-MS/MS.

3.4. Instrumentation

3.4.1 Gas Chromatography-Mass Spectrometry (GC-MS)

An Agilent 6890 GC paired with a 5973N mass selective detector (MSD) and a 7683 auto injector were used for the GC-MS analysis. Chromatographic separation was done with a HP-5ms capillary column (30 m x 0.25 mm i.d x 0.25 µm f.d) from J&W Scientific and helium as a carrier gas, flowing at a constant rate of 1.9 mL/min. The GC oven temperature program started at 70^{o}C, increased to 165^{o}C at 10^{o}C/min, and then increased to 320^{o}C at 30^{o}C/min. The oven was held at 320^{o}C for 1 minute. One microliter of sample was injected into the inlet, which was set at 235^{o}C and 18.1 psi. The

inlet was operated in pulsed splitless mode. A pulse pressure of 40 psi was used for 1 minute, and a purge flow of 50 mL/min was used at 0.75 minutes. The interface was set to 280°C. The MS quad and source were set at 150°C and 230°C respectively. The MS was operated in electron ionization mode at 70 eV. The MS was set to take full spectra scans with a range of 40-400 amu. Solvent delay was set at 1.2 minutes, and the total run time was 15.67 minutes.

GC-MS peak identities were assigned based on comparison library searches. Agilent's ChemStation software (ChemStation Software, Agilent Technologies, Santa Clara, CA), operated in Drug Analysis Mode, was used for comparison library searches. A peak was first searched against the SWGDRUG library. If no hits returned an 80% match quality than the search moved to the NIST129K library. If a match quality of 80% was still not reached, the search moved to the WILEY275 library. If the WILEY275 library did not return a search result with at least an 80% match quality, the compound was considered to be currently unidentified.

3.4.2 Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS)

Shimadzu UFLC pumps paired with an Applied Biosystems Sciex 4000 Q Trap MS/MS was used for the LC-MS/MS analysis. Chromatographic separation was achieved with a Raptor Biphenyl 2.7 µm column (50 x 2.1 mm) with a Raptor Biphenyl 2.7 µm guard cartridge (5 x 3.0 mm) attached to it, both from Restek (Restek Corporation, Bellefonte, PA). Mobile phase A consisted of 2 mM ammonium formate and 0.1% formic acid in water, while mobile phase B consisted of 2 mM ammonium formate and 0.1% formic acid in methanol. The LC pumps had a total flow rate of 0.700 mL/min. Mobile phase B concentration was held at 7.2% for 3.5 minutes, increased to

35% for 1 minute, lowered to 7.2% for 0.25 minutes, increased to 100% for 0.5 minutes, and then lowered to 7.2% for 1.75 minutes, for a total run time of 7 minutes. All changes in mobile phase B concentrations were set to immediately occur with no ramp. Injections were set at 20 µL. The oven was set at 30°C. Table 1 shows the ion transitions and LC-MS/MS instrument parameters for the 5 compounds of interest and the 4 internal standards used during this research.

Table 1. LC-MS/MS ion transitions and analytical parameters for compounds of interest.

Compound[a,b]	Q1 Mass (Da)[c]	Q3 Mass (Da)[d]	DP (volts)[e]	CE (volts)[e]	CXP (volts)[e]
Methamphetamine	150.100	91.000	56.000	25.000	14.000
	150.100	119.000	56.000	15.000	4.000
Methamphetamine-D5	155.000	91.100	60.000	20.000	4.000
Pseudoephedrine	166.180	148.024	41.000	15.000	6.000
	166.180	90.961	41.000	43.000	12.000
Pseudoephedrine-D3	169.200	151.040	26.000	21.000	26.000
Ephedrine	166.108	117.085	41.000	27.000	18.000
	166.108	114.796	41.000	35.000	18.000
Ephedrine-D3	168.980	116.999	31.000	29.000	6.000
Amphetamine	136.200	119.000	36.000	13.000	18.000
	136.200	91.000	36.000	25.000	14.000
Amphetamine-D6	142.100	125.100	41.000	13.000	6.000
CMP	152.163	79.114	41.000	27.000	12.000
	152.163	77.071	41.000	45.000	0.000

[a] Target analytes Methamphetamine, Pseudoephedrine, Ephedrine, Amphetamine, and CMP were identified using two mass ion fragments each.
[b] Internal standards Methamphetamine-d_5, Pseudoephedrine-d_3, Ephedrine-d_3, and Amphetamine-d_6 were identified using one mass ion fragment each.
[c] The values listed in column "Q1 Mass" are the molecular masses of each compound measured in Daltons.
[d] The values listed in column "Q3 Mass" are unique fragment ion masses measured in Daltons.
[e] The columns labeled "DP", "CE", and "CXP" refer to the voltages utilized for declustering potential, collision energy, and collision energy speed, respectively.

LC-MS/MS methods were built and chromatograms were observed in Analyst Software (Analyst Software, AB Sciex LLC, Framingham, MA). Compound quantitation was done in MultiQuant Software (MultiQuant Software, AB Sciex LLC, Framingham, MA).

3.5 Statistical Analysis

Statistical analyses of quantitated compounds were done in Microsoft Excel (Microsoft Office, Microsoft Corporation, Redmond, WA). These analyses were comprised of two-tailed t-tests to determine if any statistically significant differences were observed between the amount of methamphetamine, CMP, pseudoephedrine, or ephedrine found in each fraction of the ether cooks as compared to the camp fuel cooks.

3.6 Summary

Methamphetamine was produced in a laboratory setting using a method similar to those used on the street. The One Pot meth cook was separated into three components, all of which were analyzed in the laboratory using GC-MS and LC-MS/MS. Instrumental analysis was used to check for precursors, products, and unique by-products of the One Pot meth cook, which has not been done before. For GC-MS, if a compound was observed in more than one sample and it was not observed in a blank, it was considered to be a true peak and was thus considered to be a by-product of the One Pot meth cook. For LC-MS/MS, if a compound of interest was able to be quantitated above the lower limit of detection (LLOD), then it was considered to be a true peak and was said to be present in that sample.

CHAPTER IV

FINDINGS

4.1. Post-Salt Solvent

Figure 5 Figure 6 respectively show the overlain GC-MS chromatograms of the three ether cook post-salt solvents and the three camp fuel cook post-salt solvents. The reproducibility of each cook can be observed by the similarities in the chromatograms. GC-MS identified methamphetamine (RT=5.068), pseudoephedrine (RT=7.551), and ephedrine (RT=7.589) by library search and CMP (RT=5.566) by retention time and the presence of CMP's major ions at m/z 91, 77, and 58 for all the ether post-salt solvents. Also of interest, butylated hydroxytoluene (BHT) (RT=9.278) was identified by library search in the ether post-salt solvents. In the camp fuel post-salt solvents, GC-MS was only able to detect pseudoephedrine in C1 and C2, and ephedrine in C3. It should be noted that hydrocarbon chains and methylated cyclohexane compounds were not considered to be by-products on the One Pot cooks as they were found to be present in blank ether and blank camp fuel samples. Table 2 summarizes the compounds identified by GC-MS in all post-salt solvent samples.

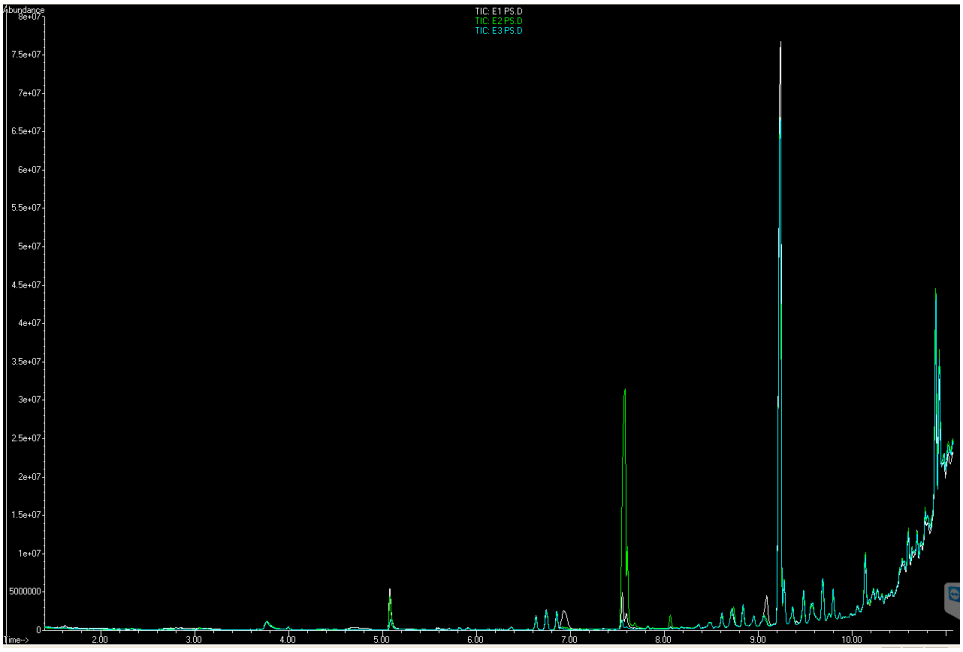

Figure 5. Overlay of E1 (white), E2 (green), and E3 (blue) post-salt solvent chromatograms with methamphetamine (RT=5.07), pseudoephedrine (RT=7.55), and ephedrine (RT=7.59) peaks visible.

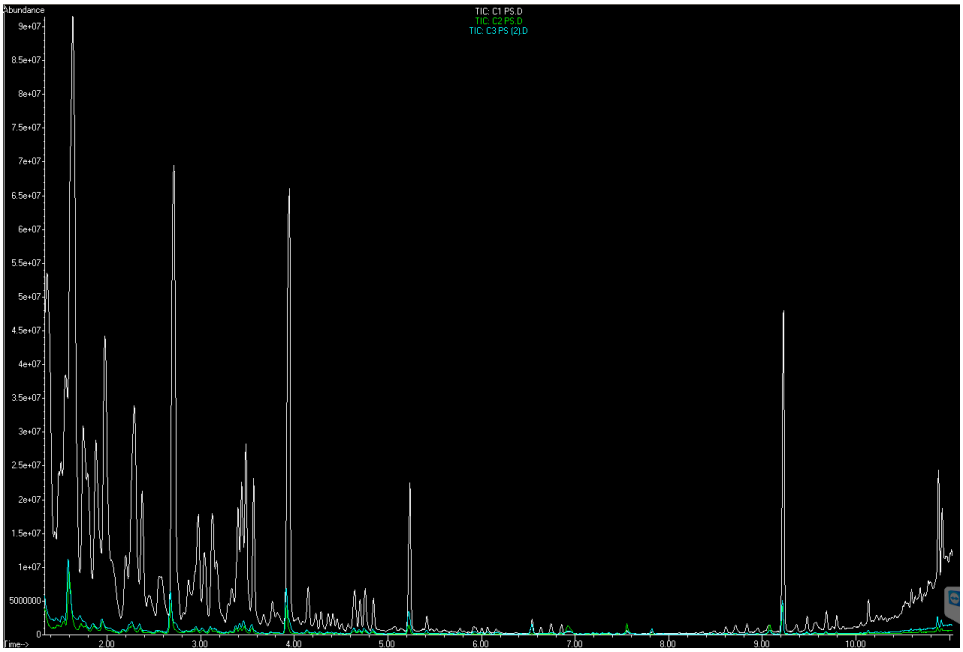

Figure 6. Overlay of C1 (white), C2 (green), and C3 (blue) post-salt solvent chromatograms.

Table 2. Summary of notable compounds identified through GC-MS analysis of each post-salt solvent.

Compound	E1	E2	E3	C1	C2	C3
Methamphetamine	X	X	X			
CMP	X	X	X			
Pseudoephedrine	X	X	X	X	X	
Ephedrine	X	X	X			X
BHT	X	X	X			

LC-MS/MS identified and quantified methamphetamine, CMP, pseudoephedrine, and ephedrine for all ether post-salt solvents. Methamphetamine, pseudoephedrine, and ephedrine were identified and quantified for all Camp fuel post-salt solvents. The quantification results are summarized in Table 3. The percentage of methamphetamine, CMP, pseudoephedrine, and ephedrine was calculated to compare the amounts of product and precursor left behind from the meth cooks. These percentages are summarized in Table 4. Percentages were calculated by using the concentrations of each chemical as determined by LC-MS/MS. The two-tailed t-tests comparing the quantitative results of the four compounds of interest in ether versus camp fuel post-salt solvents are summarized in Table 5.

Table 3. LC-MS/MS quantification results for post-salt solvents. All concentrations given in ng/mL.

Compound	E1	E2	E3	C1	C2	C3
Methamphetamine	60,600	50,600	29,700	630	20.0	808
CMP	5,800	4,030	3,420	-	-	-
Pseudoephedrine	7,370	8,630	2,900	185	6.00	424
Ephedrine	5,530	5,070	4,090	327	5.00	272

Table 4. Percent of methamphetamine, CMP, pseudoephedrine, and ephedrine found in each post-salt solvent with LC-MS/MS.

Compound	E1	E2	E3	C1	C2	C3
Methamphetamine	76.4	74.1	74.0	55.2	64.5	53.7
CMP	7.31	5.9	8.54	0.0	0.0	0.0
Pseudoephedrine	9.30	12.6	7.23	16.2	19.8	28.2
Ephedrine	6.98	7.41	10.2	28.7	15.7	18.1

Table 5. Two-tailed t-test analyses of the post-salt solvents.

	Mean Concentrations		
	Ether	Camp Fuel	p-value (two-tailed)
Post-Salt Solvent (ng/mL)			
Methamphetamine	46957	486	0.007**
CMP	4413	0.00	0.003**
Pseudoephedrine	6297	205	0.025*
Ephedrine	4896	201	0.000***

*p-values<0.05 signifying a statistical difference between the concentrations observed in the ether cooks and the camp fuel cooks.
**p-values<0.01 signifying a high statistical difference between the concentrations observed in the ether cooks and the camp fuel cooks.
***p-values<0.001 signifying an extreme statistical difference between the concentrations observed in the ether cooks and the camp fuel cooks.

4.2 Sludge

Figure 7Figure 8 respectively show the overlain GC-MS chromatograms of the three ether sludge samples and the three camp fuel sludge samples. GC-MS identified methamphetamine (RT=5.089) by library search for all six sludge samples and CMP (RT=5.562) by retention time and the presence of CMP's major ions at m/z 91, 77, and 58 for all three ether sludge samples. Pseudoephedrine and ephedrine were present in all six sludge samples, but their peaks were unable to be separated with chromatography. They were treated as a single peak, labeled "Ephedrine (Total)" and their RT was determined by the time the max height of the combined peaks was reached. Of interest,

1,2-dimethyl-3-phenylaziridine (RT=5.338) was identified by library search in all six sludge samples. An unknown compound (RT=8.121) was also observed in all sludge samples. The mass spectrum of this unknown compound contained a large peak at m/z=58 and a smaller peak at m/z=77. Figure 9 contains the chromatogram and mass spectrum of this unknown compound. Table 6 summarizes the notable compounds found in the sludge samples.

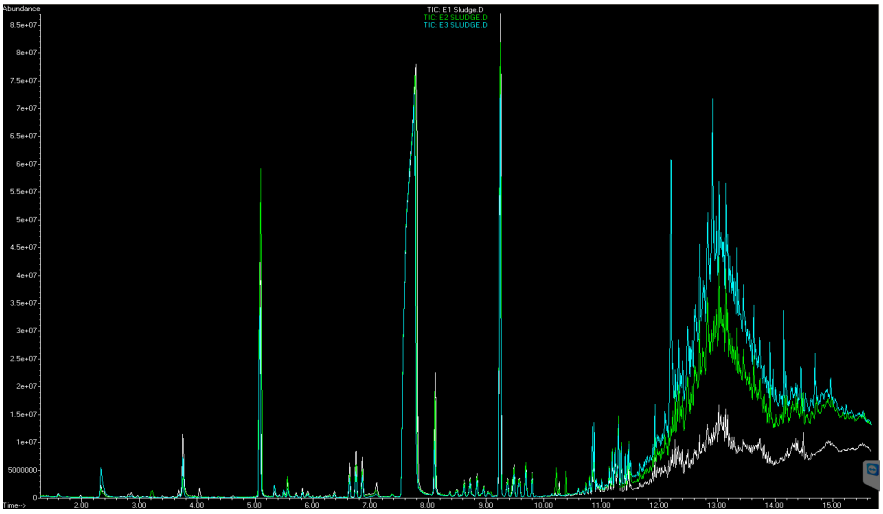

Figure 7. Overlay of E1 (white), E2 (green), and E3 (blue) wet sludge chromatograms with methamphetamine (RT=5.089), CMP (RT=5.562), and the ephedrine (total) (RT=7.775) peaks visible.

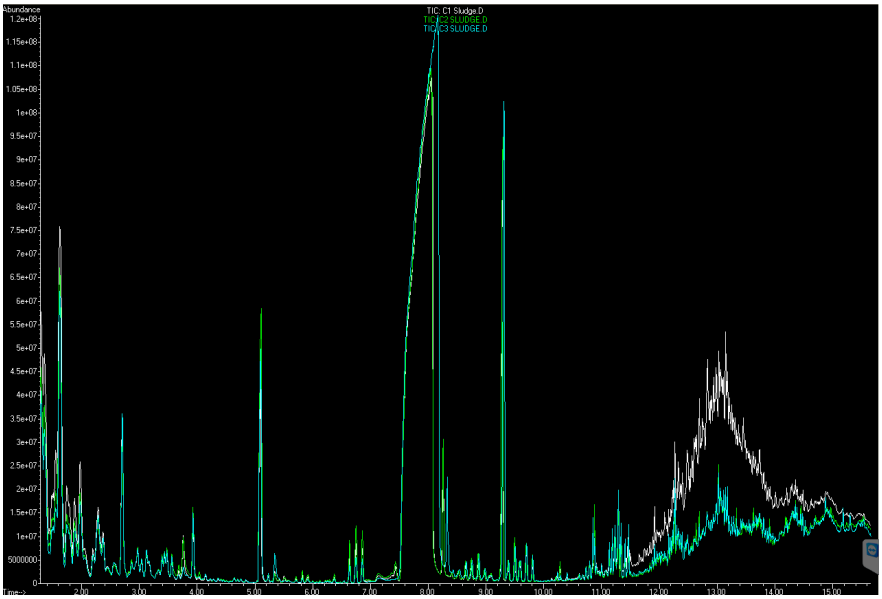

Figure 8. Overlay of C1 (white), C2 (green), and C3 (blue) wet sludge chromatograms with the methamphetamine (RT=5.098) and the ephedrine (total) (RT=8.171) peaks visible.

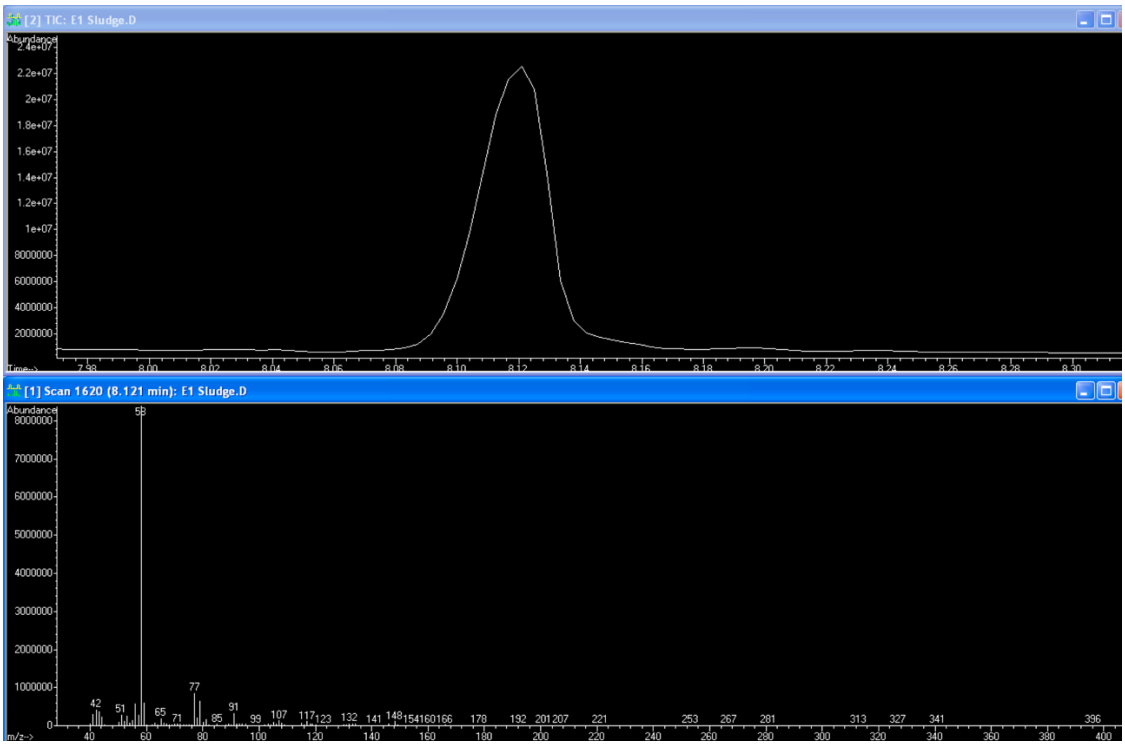

Figure 9. Chromatogram and mass spectrum of the unknown compound found in all six sludge samples.

Table 6. Summary of notable compounds identified through GC-MS analysis of each sludge sample.

Compound	E1	E2	E3	C1	C2	C3
Methamphetamine	X	X	X	X	X	X
CMP	X	X	X			
Ephedrine (Total)	X	X	X	X	X	X
1,2-dimethyl-3-phenylaziridine	X	X	X	X	X	X
Unknown Compound	X	X	X	X	X	X

LC-MS/MS identified and quantified methamphetamine, CMP, pseudoephedrine, and ephedrine for all ether sludge samples. Methamphetamine, pseudoephedrine, and ephedrine were identified and quantified for all camp fuel sludge samples. The quantification results are summarized in Table 7. The percentage of methamphetamine, CMP, pseudoephedrine, and ephedrine found in each sludge sample is summarized in

Table 8. The two-tailed t-tests comparing the quantitative results of the four compounds of interest in ether versus camp fuel sludge samples are summarized in Table 9.

Table 7. LC-MS/MS quantification results for sludge samples. All concentrations given in mg/mL.

Compound	E1	E2	E3	C1	C2	C3
Methamphetamine	4.46	8.78	2.66	0.31	1.14	0.78
CMP	0.31	0.35	0.19	-	-	-
Pseudoephedrine	0.94	0.57	0.82	0.39	0.45	0.61
Ephedrine	4.30	2.46	3.05	1.86	2.15	3.24

Table 8. Percent of methamphetamine, CMP, pseudoephedrine, and ephedrine found in each sludge sample with LC-MS/MS.

Compound	E1	E2	E3	C1	C2	C3
Methamphetamine	44.6	72.3	39.6	12.0	30.5	16.9
CMP	3.07	2.89	2.8	0.00	0.00	0.00
Pseudoephedrine	9.35	4.66	12.2	15.3	12.2	13.1
Ephedrine	43.0	20.2	45.4	72.7	57.3	70.0

Table 9. Two-tailed t-test analyses of the sludge samples.

	Mean Concentrations		
	Ether	Camp Fuel	p-value (two-tailed)
Sludge (mg/mL)			
Methamphetamine	5.30	0.74	0.068
CMP	0.28	0.00	0.004**
Pseudoephedrine	0.77	0.48	0.084
Ephedrine	3.27	2.41	0.283

*p-values<0.05 signifying a statistical difference between the concentrations observed in the ether cooks and the camp fuel cooks.
**p-values<0.01 signifying a high statistical difference between the concentrations observed in the ether cooks and the camp fuel cooks.
***p-values<0.001 signifying an extreme statistical difference between the concentrations observed in the ether cooks and the camp fuel cooks.

4.3 Product Salts

Figure 10Figure 11 respectively show the overlain GC-MS chromatograms of the three product salts synthesized with ether and the three product salts synthesized with camp fuel. GC-MS identified methamphetamine and ephedrine by library search for all six product salt samples. CMP was identified by retention time and the presence of its major ions at m/z 91, 77, and 58 for all three ether product salts. Pseudoephedrine was identified by library search in all six camp fuel product salts. Table 10 summarizes the notable compounds found in the product salts synthesized during the One Pot cooks.

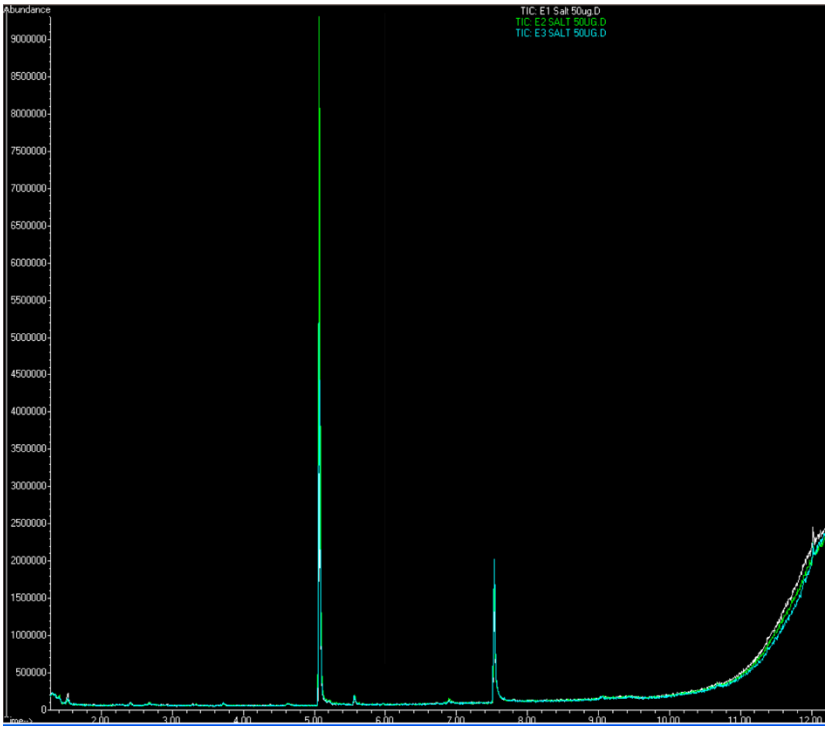

Figure 10. Overlay of E1 (white), E2 (green), and E3 (blue) product salt chromatograms with methamphetamine (RT=5.072), CMP (RT=5.566), and the ephedrine (RT=7.614) peaks visible.

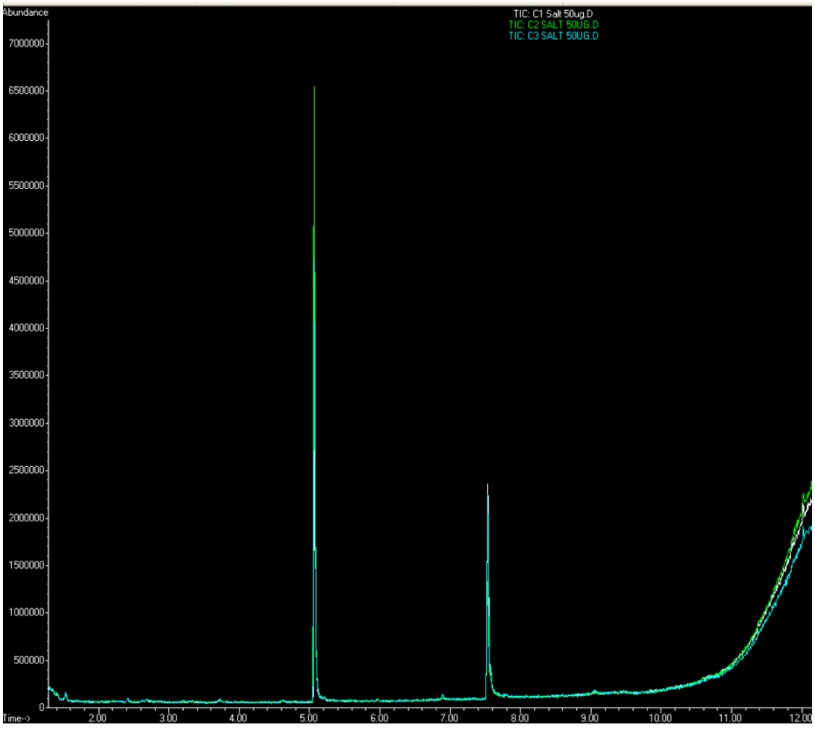

Figure 11. Overlay of C1 (white), C2 (green), and C3 (blue) product salt chromatograms with methamphetamine (RT=5.068), and the combination pseudoephedrine (RT=7.534) / ephedrine (RT=7.576) peaks visible.

Table 10. Summary of notable compounds identified through GC-MS analysis of each product salt.

Compound	E1	E2	E3	C1	C2	C3
Methamphetamine	X	X	X	X	X	X
CMP	X	X	X			
Pseudoephedrine				X	X	X
Ephedrine	X	X	X	X	X	X

LC-MS/MS identified and quantified methamphetamine, CMP, pseudoephedrine, and ephedrine for all six product salts. The quantification results are summarized in Table 11. The percentage of methamphetamine, CMP, pseudoephedrine, and ephedrine found in each product salt is summarized in Table 12. The two-tailed t-tests comparing the quantitative results of the four compounds of interest in ether versus camp fuel product salts are summarized in Table 13.

Table 11. LC-MS/MS quantification results for the product salts. All concentrations given in ng/mL.

Compound	E1	E2	E3	C1	C2	C3
Methamphetamine	21.8	56.3	35.9	23.1	35.6	25.8
CMP	1.62	2.19	2.28	0.22*	0.24*	0.16*
Pseudoephedrine	8.60	8.39	12.0	14.7	10.4	14.2
Ephedrine	36.9	44.2	52.1	59.8	33.4	49.7

*Concentration is below the LOQ of 0.5 ng/mL.

Table 12. Percent of methamphetamine, CMP, pseudoephedrine, and ephedrine found in each product salt with LC-MS/MS.

Compound	E1	E2	E3	C1	C2	C3
Methamphetamine	31.6	50.7	35.1	23.6	44.7	28.7
CMP	2.35	1.97	2.23	0.23	0.30	0.18
Pseudoephedrine	12.5	7.56	11.7	15.0	13.1	15.8
Ephedrine	53.6	39.8	50.9	61.2	41.9	55.3

Table 13. Two-tailed t-test analyses of the product salts.

	Mean Concentrations		
	Ether	Camp Fuel	p-value (two-tailed)
Salts (ng/mL)			
Methamphetamine	37.96	28.16	0.412
CMP	2.03	0.00	0.001**
Pseudoephedrine	9.66	13.08	0.126
Ephedrine	44.39	47.65	0.732

*p-values<0.05 signifying a statistical difference between the concentrations observed in the ether cooks and the camp fuel cooks.
**p-values<0.01 signifying a high statistical difference between the concentrations observed in the ether cooks and the camp fuel cooks.
***p-values<0.001 signifying an extreme statistical difference between the concentrations observed in the ether cooks and the camp fuel cooks.

CHAPTER V

CONCLUSIONS

5.1 Results Discussion

5.1.1 Post-Salt Solvent

GC-MS analysis of the post-salt solvents from the ether and camp fuel One Pot meth cooks revealed several pieces of information. First, the use of different solvents in meth cooks can drastically change the by-products produced in this liquid waste. CMP, commonly thought of as the primary impurity in the Birch reduction method of methamphetamine synthesis, was clearly seen on the GC-MS chromatograms of the ether post-salt solvents, but was undetectable on the camp fuel chromatograms, even when the chromatograms were subjected to extracted ion chromatogram searches.[33] These findings were verified by LC-MS/MS analysis, which found CMP in the ether post-salt solvents in the range of 3000-6000 ng/mL, but was unable to identify CMP in the camp fuel post-salt solvents.

GC-MS analysis was also unable to identify methamphetamine in the camp fuel post-salt solvents, but LC-MS/MS identified it and quantified it at a hundred-fold lower concentration then what was found in the ether post-salt solvents. GC-MS was only able to detect pseudoephedrine in the C1 and C2 post-salt solvent and ephedrine in the C3 post-salt solvent. LC-MS/MS was able to detect both pseudoephedrine and ephedrine in

all three camp fuel post-salt solvents at a ten-fold lower concentration then what was observed in the ether cooks. In all six post-salt solvents, methamphetamine was found at a higher concentration than any of the other compounds of interest.

Statistical analysis of the concentrations of the four compounds of interest in the ether post-salt solvents versus the camp fuel post-salt solvents revealed a significant difference of all four compounds between the two cook solvents. Pseudoephedrine had a p-value of 0.025, signifying a significant difference between the amount of pseudoephedrine found in the ether post-salt solvents and the camp fuel post-salt solvents. Methamphetamine and CMP had p-values of 0.007 and 0.003 respectively, signifying a highly significant difference between the concentrations of these two drugs in the ether post-salt solvents when compared to the camp fuel post-salt solvents. Ephedrine had a p-value of 0.000, signifying an extremely significant difference between the concentration of ephedrine in the ether post-salt solvents and the camp fuel post-salt solvents. These statistically significant differences in concentrations of the 4 compounds of interest in the ether post-salt solvents and the camp fuel post-salt solvents may be of interest for future research.

Besides the four compounds of interest, butylated hydroxytoluene (BHT) was observed in all 3 ether post-salt solvents by GC-MS. This by-product has been previously reported as an impurity in seized methamphetamine samples.[39] BHT is commonly added to diethyl ether to prevent the formation of peroxides and improve the stability and shelf life of the solvent.[40]

5.1.2 Sludge

The use of different solvents also led to different amounts of product being left in the sludge of these reactions. Methamphetamine was found in all 6 sludge samples with GC-MS and LC-MS/MS. While found in the ether sludge samples at a concentration four-times greater than what was observed in the camp fuel sludge samples, there was no significant difference (p-value=0.068) in the concentrations of methamphetamine found in the sludge of either solvent type. The concentrations of pseudoephedrine and ephedrine observed in the three ether sludge samples using LC-MS/MS also was not significantly different from the concentrations observed in the camp fuel sludge samples. As with the post-salt solvents, CMP was only found in the ether samples and it was observed with both GC-MS and LC-MS/MS.

GC-MS analysis identified 2 by-products in the sludge samples. The first by-product, 1,2-dimethyl-3 phenylaziridine (RT=5.338), was identified by a library search. This by-product was observed in all six sludge samples, and it is primarily considered a by-product of the Red-P method of methamphetamine production, though it can be produced anytime an acid is allowed to dehydrate ephedrine or pseudoephedrine.[34] The second by-product observed during GC-MS analysis of the sludge samples was found at RT=8.121, and its identity is unknown. Its mass spectrum, shown in Figure 9 along with its chromatographic peak, had a strong m/z peak at 58 and a smaller one at 77. Library search results returned multiple hits, all of which matched by more than 70 percent. These matches included pseudoephedrine, ephedrine, 3-ethoyx methamphetamine, and benzenemethanol. This compound may be of interest for further research.

5.1.3 Product Salts

LC-MS/MS analysis of the product salts from both cook types revealed similar that ether product salts and camp fuel product salts were only different in that ether product salts contained CMP while camp fuel product salts did not. The amounts of methamphetamine, pseudoephedrine, and ephedrine present in the product salts were all similar for both cook types. GC-MS analysis of the product salts provided no additional by-products to the product salts other than CMP.

While instrumental analysis showed very little difference between the ether product salts and the camp fuel product salts, physical observations of these product salts showed two major differences. The first difference was in the color of the product salts. The ether product salts were always pure white. The camp fuel product salts varied in color from robin's egg blue to cotton candy pink. A side-by-side image of these salts is shown in Figure 12. The color of the camp fuel product salts is believed to come from retention of the camp fuel during the One Pot meth cook, as the camp fuel itself can vary in color.

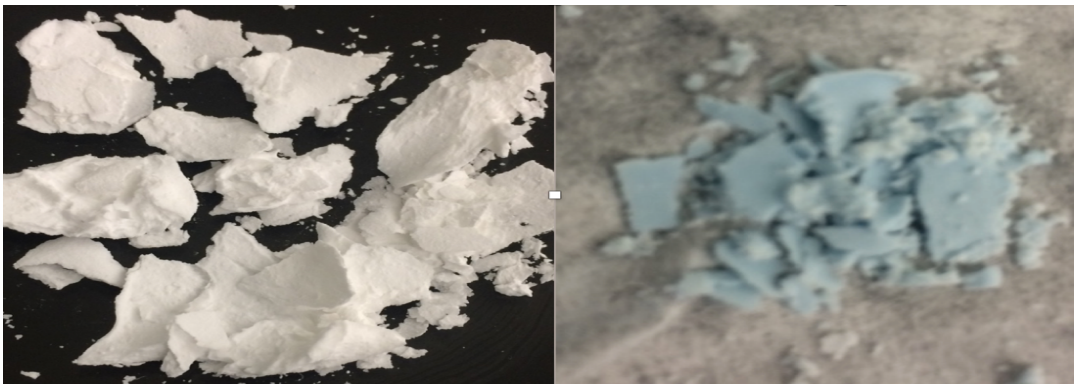

Figure 12. Comparison of the product salts synthesized during the ether cook (left) and the camp fuel cook (right). The blue color of the camp fuel product salt is believed to come from the retention of some of the camp fuel used during the meth cook.

The other physical difference observed between the ether product salts and the camp fuel product salts is their weights. The ether One Pot cooks resulted in an average product salt mass of 1.257 g while the camp fuel One Pot cooks resulted in an average product salt mass of 0.166 g. It is believed that this difference in mass stems from ether's ability to solvate water. During the One Pot meth cook, water is added to the reaction to catalyze it. When methamphetamine is in its freebase form, it will stay in the organic solvent and not move to the water. However, to get meth out of solution, HCl gas is bubbled into the organic solvent. The addition of this acid gas causes freebase meth to convert to methamphetamine-HCl, which is no longer soluble in organic solvents but is soluble in aqueous solvents. In ether One Pot meth cooks, the methamphetamine-HCl has to compete with diethyl ether to dissolve into the water that is present in the cook. Since there are far more ether molecules than meth-HCl molecules, very little of the meth stays in solution and most of it is filtered out. In camp fuel One Pot cooks, methamphetamine-HCl has very little competition for the water, as camp fuel is very hydrophobic and doesn't mix well with water. This means that some meth-HCl may be left behind in the small amount of water that is present in the post-salt solvent. If the amount of water left in the post-salt solvent is small enough, the separation between it and the camp fuel may not be noticeable, thus it was not analyzed with LC-MS/MS. This difference in mass may be of interest for further research.

5.2 Conclusion

One Pot methamphetamine cooks were performed in a laboratory environment using two different organic solvents for the purpose of chemical characterization via gas chromatography-mass spectrometry and liquid chromatography-tandem mass

spectrometry. One Pot meth cooks were divided into three fractions for characterization: post-salt solvent, sludge, and product salts. All fractions were analyzed with GC-MS in order to identify any chemical by-products that were present in the fraction. After GC-MS analysis, LC-MS/MS was used to quantify four drugs of interest: methamphetamine, 1-(1,4-cyclohexadienyl)-2-methylaminopropane (CMP), pseudoephedrine and ephedrine.

The goal of this study was to answer three research questions: 1.) Does waste generated from a One Pot methamphetamine lab contain chemical by-products unique to this cook method? 2.) Are any by-products observed in One Pot methamphetamine waste currently unidentified compounds? 3.) Does the type of solvent used during a One Pot methamphetamine cook make a difference in the by-products observed in the cook waste? As far as unique chemical by-products, the One Pot method of methamphetamine production did not contain any by-products that have not already been reported in previous methods of methamphetamine production. The three major by-products observed within the One Pot cooks were CMP, 1,2-dimethyl-3-phenylaziridine, and an unknown pseudoephedrine-like compound. CMP and the unknown pseudoephedrine-like compound have both previously been identified in Birch reduction meth cooks and 1,2-dimethyl-3-phenylaziridine has previously been identified in Red-P meth cooks.[4,34] Currently the identity of the pseudoephedrine-like by-product is unknown. While this compound was previously reported as being present in methamphetamine produced via the Birch reduction method, its actual chemical structure was never determined, making this compound something of interest for further research.[4] Production of the preceding by-products was determined to be altered based on the organic solvent used during the One Pot meth cooks. It was shown that CMP production was hindered when camp fuel

was used instead of ether. It was also shown that a smaller mass of product salts were obtained from cooks that used camp fuel instead of ether as a solvent. While it is currently unknown why the solvent affects the One Pot cooks in the way that it does, this is another area of interest for further research.

Now that some of the by-products produced in One Pot methamphetamine labs have been identified, further research can be done on these compounds to determine if more advanced personal protective equipment is needed to protect first responders and law enforcement officers. The identification of these by-products may also be useful in developing new ways to identify One Pot meth labs so they may be seized. Clandestine methamphetamine manufacture is hazardous, and proper characterization of the methods is necessary to identify laboratories and mitigate their public health impact.

REFERENCES

1. Kleinschmidt K. What is in a meth lab? http://www.acmt.net/_Library/2015_Meth_Course_Billings_MT/1_6_What_is_in_a_Meth_Lab_ppt.pdf. Accessed November 8, 2016.

2. Scherbenske JR. Methamphetamine and the CMEA. July 2014. https://www.deadiversion.usdoj.gov/mtgs/pharm_awareness/conf_2014/july_2014/scherbenske.pdf. Accessed November 8, 2016.

3. Kunalan V, Kerr WJ, Daéid NN. Investigation of the reaction impurities associated with methylamphetamine synthesized using the Nagai method. *Anal Chem*. 2012;84:5744-5752. doi:10.1021/ac3009302.

4. Kunalan V, Kerr WJ, NicDaéid N. Clarification of route specific impurities found in methylamphetamine synthesised using the Birch method. *Forensic Sci Int*. 2012;223(1–3):321-329. doi:10.1016/j.forsciint.2012.10.008.

5. United States Environmental Protection Agency. Voluntary guidelines for methamphetamine laboratory cleanup. https://www.epa.gov/sites/production/files/documents/meth_lab_guidelines.pdf. Revised March 2013. Accessed September 11, 2016.

6. Mach PM, McBride EM, Sasiene ZJ, et al. Vehicle-mounted portable mass spectrometry system for the covert detection via spatial analysis of clandestine methamphetamine laboratories. *Anal Chem*. 2015;87(22):11501-11508. doi:10.1021/acs.analchem.5b03269.

7. Gardner TJ, Anderson TM. Drug abuse and alcohol related crimes. In: Gardner TJ, Anderson TM. *Criminal Law*. 12th ed. Stamford, CT: Cengage Learning; 2014:444-471. https://books.google.com/books?id=vOIbCgAAQBAJ. Accessed August 25, 2016.

8. Volkow ND. Methamphetamine. National Institute on Drug Abuse Web site. https://www.drugabuse.gov/publications/research-reports/methamphetamine/letter-director. Published September 2013. Accessed April 19, 2016.

9. Methamphetamine lab incidents. DEA Web site. https://www.dea.gov/resource-center/meth-lab-maps.shtml. Accessed April 1, 2016.

10. Drug Enforcement Agency. National drug threat assessment summary 2015. https://www.dea.gov/docs/2015%20NDTA%20Report.pdf. Published October 2015. Accessed April 3, 2016.

11. Maxwell JC, Brecht M-L. Methamphetamine: here we go again? *Addict Behav*. 2011;36(12):1168-1173. doi:10.1016/j.addbeh.2011.07.017

12. Weisheit R, White WL. *Methamphetamine: Its History, Pharmacology and Treatment*. Center City, MN: Hazelden Publishing; 2009. https://books.google.com/books?id=YqZkw-apxVAC. Accessed April 3, 2016.

13. The truth about crystal meth. Foundation for a Drug-Free World Web site. http://www.drugfreeworld.org/drugfacts/crystalmeth/history-of-methamphetamine.html. Accessed April 3, 2016.

14. Controlled Substances Act, 21 USC §801-904 (1970).

15. Cunningham JK, Maxwell JC, Campollo O, Cunningham KI, Liu L-M, Lin H-L. Proximity to the US–Mexico border: a key to explaining geographic variation in US methamphetamine, cocaine and heroin purity. *Addiction*. 2010;105(10):1785-1798. doi:10.1111/j.1360-0443.2010.03032.x.

16. Weisheit R. Making methamphetamine. *South Rural Sociol*. 2008;23(2):78-107.

17. National Drug Intelligence Center. National drug threat assessment 2011. https://www.justice.gov/archive/ndic/pubs44/44849/44849p.pdf. Accessed April 13, 2016.

18. Phenylacetone (phenyl-2-propanone). Chemical LAND21 Web site. http://www.chemicalland21.com/specialtychem/finechem/PHENYLACETONE.htm. Accessed April 19, 2016.

19. Methylamines uses & applications. Chemours Web site. https://www.chemours.com/Methylamines/en_US/uses_apps/index.html. Accessed April 19, 2016.

20. Nordin J. More on meth lab chemicals. AristaTek Web site. http://www.aristatek.com/newsletter/0511November/TechSpeak.aspx. Published November 2005. Accessed April 10, 2016.

21. American Industrial Hygiene Association. Meth chemistry: "What's Cookin'?" https://www.aiha.org/aihce06/handouts/rt236cameron.pdf. Accessed April 14, 2016.

22. Skinner HF. Methamphetamine synthesis via HI/red phosphorous reduction of ephedrine. *Forensic Sci Int*. 1990;48(2):128-134. doi:10.1016/0379-0738(90)90104-7.

23. Comprehensive Methamphetamine Control Act of 1996, 21 USC §801-904 (1996).

24. Brzeczko AW, Leech R, Stark JG. The advent of a new pseudoephedrine product to combat methamphetamine abuse. *Am J Drug Alcohol Abuse*. 2013;39(5):284-290. doi:10.3109/00952990.2013.821476.

25. Oz M. Birch reduction of pseudoephedrine to methamphetamine. Erowid Web site. https://erowid.org/archive/rhodium/chemistry/birch.pseudo.html. Accessed April 2, 2016.

26. Combat Methamphetamine Epidemic Act of 2005, 21 USC §801-904 (2006).

27. Street D. An investigator's guide to the methamphetamine lab. *Police*. 2000:1-2. http://www.policemag.com/channel/patrol/articles/2000/12/an-investigator-s-guide-to-the-methamphetamine-lab.aspx. Accessed September 9, 2016.

28. Oklahoma Bureau of Narcotics and Dangerous Drugs. Guidelines for cleaning up former meth labs. http://www.deq.state.ok.us/lpdnew/MethLabs/meth.htm. Accessed September 11, 2016.

29. Bartos L. Meth production is toxic to communities (methtoxic). University of Nebraska-Lincoln Web site. http://lancaster.unl.edu/family/methtoxic.shtml. Published July 2005. Accessed April 15, 2016.

30. Huerta-Fontela M, Galceran MT, Martin-Alonso J, Ventura F. Occurrence of psychoactive stimulatory drugs in wastewaters in north-eastern Spain. *Sci Total Environ*. 2008;397(1–3):31-40. doi:10.1016/j.scitotenv.2008.02.057.

31. Mwenesongole E, Gautam L, Hall S, Waterhouse J, Cole MD. Simultaneous detection of controlled substances in waste water. *Anal Methods*. 2013;5:3248-3254. doi:10.1039/C3AY40655E.

32. Triplett JS, Hatfield JA, Kaeff TL, Ramsey CR, Robinson SD, Standifer AF. Raman spectroscopy as a simple, rapid, nondestructive screening test for methamphetamine in clandestine laboratory liquids. *J Forensic Sci*. 2013;58(6):1607-1614. doi:10.1111/1556-4029.12213.

33. Person EC, Meyer JA, Vyvyan JR. Structural determination of the principal byproduct of the lithium-ammonia reduction method of methamphetamine manufacture. *J Forensic Sci*. 2005;50(1):1-9. doi:10.1520/JFS2003389.

34. Stojanovska N, Fu S, Tahtouh M, Kelly T, Beavis A, Kirkbride KP. A review of impurity profiling and synthetic route of manufacture of methylamphetamine, 3,4-methylenedioxymethylamphetamine, amphetamine, dimethylamphetamine and p-methoxyamphetamine. *Forensic Sci Int*. 2013;224(1–3):8-26. doi:10.1016/j.forsciint.2012.10.040.

35. O'Leary AE, Hall SE, Vircks KE, Mulligan CC. Monitoring the clandestine synthesis of methamphetamine in real-time with ambient sampling, portable mass spectrometry. *Anal Methods*. 2015;7:7156-7163. doi:10.1039/c5ay00511f.

36. Jurkanin TJ. Law enforcement response to methamphetamine. *Law Enforc Exec Forum*. 2003. https://www.iletsbeiforumjournal.com/images/Issues/FreeIssues/ILEEF%202003-3.4.pdf. Accessed September 18, 2016.

37. Waxman OB. Meth lab found in Walmart bathroom. *Time*. March 13, 2015. http://time.com/3744599/walmart-restroom-meth-lab/. Accessed April 21, 2016.

38. New "shake-and-bake" method for making crystal meth gets around drug laws but is no less dangerous. *New York Daily News*. August 25, 2009. http://www.nydailynews.com/life-style/health/new-shake-and-bake-method-making-crystal-meth-drug-laws-no-dangerous-article-1.397428. Accessed April 1, 2016.

39. Koester CJ, Andresen BD, Grant PM. Optimum methamphetamine profiling with sample preparation by solid-phase microextraction. *J Forensic Sci.* 2002;47(5):1002-1007. doi:10.1520/JFS15505J.

40. Solvent Stabilizer Systems. Sigma-Aldrich Web site. http://www.sigmaaldrich.com/chemistry/solvents/learning-center/stabilizer-systems.html. Accessed January 26, 2017.

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